Occurrence of Bacterial Spot and Bacterial Canker of Tomato in the Russian Federation

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Abstract

Bacterial spot of tomato, caused in Russia by *X. vesicatoria* (Group B of *X. campestris* pv. *vesicatoria*) and *X. gardneri* (Group D) (Jones et al., 2004) and bacterial canker (*C. michiganensis* subsp. *michiganensis*) have become important diseases of tomato in the Russian Federation. Over 100 samples of tomato plants showing symptoms of bacterial canker or black spots were collected in the Moscow, Pskov, Tver, and Saratov regions, and the Republic of Tatarstan from June 2006 through June 2007. Symptoms on most plants consisted of black spots with yellow halos and wilting lesions surrounded by yellowing, normally with brown veins. Sixty-seven yellow-pigmented strains isolated from lesions were compared by phytopathologic, biochemical and molecular tests to reference strains of *X. vesicatoria* (NCPPB 422T, XV 153 and XV 938), *X. gardneri* (XV GA2), and *C. michiganensis* subsp. *michiganensis* (5213). Only a single pathogen was recovered from 24 samples for xanthomonads and from 38 samples for clavibacteria. Forty of 102 samples exhibited combined infection by both pathogens. Unexpectedly, some xanthomonads of both groups B and D were isolated together with *C. michiganensis* subsp. *michiganensis* from leaves with wilting symptoms only.

INTRODUCTION

Bacterial spot of tomato, caused in Russia by *X. vesicatoria* (Group B – Xv) and *X. gardneri* (Group D - Xg) (Jones et al., 2004) and bacterial canker (*C. michiganensis* subsp. *michiganensis* - Cmm) are important diseases of tomato in the Russian Federation. Symptoms of bacterial canker were observed in glasshouses in the Moscow, Tver, and Pskov regions and Tatarstan, at the same places and time as symptoms of bacterial spo. Moreover, both pathogens (Cmm and xanthomonads) were repeatedly isolated from the same diseased plants with symptoms of wilting, leaf blight, and leaf spots.

MATERIALS AND METHODS

Over 100 samples of diseased tomato plants with either bacterial canker or black leaf spot symptoms were collected in glasshouses in several regions of the Russian Federation. The samples were evaluated for mixed infection and the strains retained for pathogenicity, biochemical, and genetic tests.

Yellow-pigmented *Clavibacter*- and *Xanthomonas*-like bacteria were isolated from diseased plants in several regions on semi-selective media: mSCM for Cmm (Schaad et al., 2001) and KA (agar 2%, Tween 80 1%, glucose 5%, gentamycin 20 mg/l) for Xv/Xg (Dzhalilov et al., 2007). Bacteria were purified by several passages on YDC medium, and stored for further analysis. For inoculation, bacterial cultures were grown in liquid NBY overnight, and adjusted to 1 x 10^6 cfu/ml.

The hypersensitivity test (HR) for Xv/Xg was tested on geranium, plectrantus, and tobacco plant leaves; while Cmm was tested on *Mirabilis* sp. plants For host

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pathogenicity assays, seedlings of susceptible tomato cv. 'Dubok' were sprayed with Xv orXg or clipped (Cmm) with the inoculum.

To confirm the presence of Xv, Xg and/or Cmm in plant tissues, BIO-PCR analysis was done by incubation of samples washed from diseased plants on water agar for 24h at 28°C and PCR amplification with primers 743/1443 (Tsygankova et al., 2004) modified to amplify both Xv and Xg, and primers D11F/R (F 5'-GGA GAA CAG GCC CGT GA-3'; R 5'-CTC CGC GCG ATC CCC ACC-3') for Cmm (Karlov et al., 2007).

RESULTS AND DISCUSSION

Isolation on KB media recovered Xv/Xg from 57% of 102 samples, while Cmm was recovered on mSCM medium from 68% of the samples. Only 24.5% of the plants were positive for both pathogens. BIO-PCR analysis identified 15 additional samples with both pathogens, 59% more than by conventional isolation.

CONCLUSIONS

Two of four species of the former *X. campestris* pv. *vesicatoria* causing black spot disease of tomatoes - *X. vesicatoria* (Group B) and *X. gardneri* (Group D) - are present in Russia on tomato plants cultivated in glasshouses.

Both major bacterial tomato pathogens in Russia: *X. vesicatoria/X. gardneri* and *Clavibacter michiganensis* subsp. *michiganensis* are often found on tomato plants grown in glasshouses as mixed infections.

Conventional isolation on semi-selective media detects both pathogens with only in 62% of cases comparing to BIO-PCR analysis.

Bacteria of *X. vesicatoria/X. gardneri* could be present in tomato plants with bacterial canker (wilting) without typical black leaf spots symptoms.

Seed of tomato must be tested for both pathogens – *Clavibacter michiganensis* subsp. *michiganensis* and *X. vesicatoria/X. gardneri* in order to prevent disease epidemic in tomatoes under glasshouse production systems

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Tables

Table 1. Identification of *X. vesicatoria* (Xv), *X. gardneri* (Xg) and *C. michiganensis* subsp. *michiganensis* (Cmm) in diseased tomato samples by isolation on semi-selective media and BO-PCR assay.

Pathogens	Samples positive on mSCM and KB	Samples positive in BIO-PCR	Difference between isolation and BIO-PCR
Only Xv/Xg	33 (32.5%)	24 (23.5%)	9
Xv/Xg + Cmm	25 (24.5%)	40 (39.5%)	-15
Only Cmm	44 (43.5%)	38 (37.5%)	6
All Xv/Xg	58 (57%)	64 (63%)	-6
All Cmm	69 (68%)	78 (77%)	-9
Total	102 (100%)	102 (100%)	For all Cmm and Xv/Xg = -15 (-14.5%)